
A STUDY OF THE ANALGESIC, ANTIDIARRHOEAL AND ANTIMICROBIAL ACTIVITIES OF LIMNOPHILA POLYSTACHYA BENTH (SCROPHULARIACEAE) LEAF EXTRACTS

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ABSTRACT

Phytopharmacology continues to play a crucial role in the discovery and development of novel therapeutic agents, particularly from medicinal plants with traditional uses. The increasing prevalence of antibiotic resistance, along with the widespread occurrence of pain and diarrhoeal disorders, has created an urgent need for safer and more effective plant-based alternatives. This study evaluated the analgesic, antidiarrhoeal, and antimicrobial activities of ethanol leaf extracts of *Limnophila polystachya* Benth using established experimental models. Analgesic activity was assessed by the acetic acid-induced writhing test in mice, while antidiarrhoeal activity was evaluated using the castor oil-induced diarrhoea model following Thomas' method. Antimicrobial activity was determined by the disc diffusion assay against selected bacterial strains. At a dose of 500 mg/kg, the stem and leaf extracts produced significant analgesic effects by reducing writhing responses by 65.79% and 63.16%, respectively, compared with 78.07% inhibition produced by the standard drug diclofenac sodium. In the antidiarrhoeal study, the stem and leaf extracts significantly reduced diarrhoeal faeces by 50.58% and 52.87%, respectively, while lower doses also demonstrated notable inhibition. The extracts exhibited moderate antibacterial activity at concentrations of 250 and 500 µg/disc, although their efficacy was lower than ciprofloxacin (38 mm inhibition zone at 50 µg/disc). These findings indicate that *L. polystachya* possesses promising pharmacological properties and may serve as a valuable natural source for the isolation of bioactive compounds. However, further phytochemical investigations, toxicity studies, and mechanistic

evaluations are required to identify the active constituents and confirm their therapeutic potential.

KEYWORDS: *Limnophila polystachya*; Phytopharmacology; Analgesic activity; Antidiarrhoeal activity; Antimicrobial activity; Medicinal plants.

INTRODUCTION

Plant secondary metabolites are an important source of therapeutic agents and contribute significantly to healthcare worldwide. Diarrhoea, characterized by frequent bowel movements, watery stools, and abdominal pain, can lead to dehydration and electrolyte imbalance if left untreated. Analgesics relieve pain by acting on the central and peripheral nervous systems without affecting consciousness, and both central and peripheral pain models are commonly used to evaluate their efficacy. Medicinal plants have long been recognized for their antimicrobial properties and remain a rich source of compounds with antibacterial, antifungal, antiviral, and antiprotozoal activities.

Limnophila polystachya Benth, a member of the family Scrophulariaceae, is a medicinal herb traditionally used in Indian medicine to treat fever, dysentery, elephantiasis, dyspepsia, and respiratory disorders, as well as to promote lactation. Previous studies have reported its antibacterial, antifungal, antioxidant, antitumor, antimutagenic, antimycobacterial, and antineoplastic properties. However, phytochemical and pharmacological investigations of *L. polystachya* remain limited. Research on related *Limnophila* species has identified several bioactive flavonoids and essential oils, suggesting that *L. polystachya* may also contain valuable phytochemicals with significant therapeutic potential. Further phytochemical characterization and pharmacological studies are needed to identify its active constituents and validate its medicinal applications.

IMPORTANCE OF MEDICINAL PLANTS IN DRUG DISCOVERY

Medicinal plants have played a fundamental role in drug discovery and continue to serve as an invaluable source of therapeutic agents, particularly for the treatment of infectious diseases. Many plant-derived bioactive compounds are used directly as medicines, while others act as lead molecules for the development of new drugs through chemical modification or semisynthesis. Recent advances in biotechnology, genome mining, analytical techniques, microbial culture methods, and high-throughput screening have significantly improved the identification, isolation, and evaluation of natural compounds. These innovations enable

researchers to better understand the molecular mechanisms of action of plant-derived compounds and accelerate the drug development process. Growing concerns over antimicrobial resistance have renewed interest in medicinal plants as potential sources of novel antimicrobial agents. Modern drug discovery relies on interdisciplinary collaboration among experts in ethnobotany, pharmacognosy, medicinal chemistry, molecular biology, pharmacology, microbiology, and related fields. The discovery process generally involves plant collection, extraction, isolation of bioactive constituents, structural characterization, and biological evaluation. When combined with computational modeling, synthetic chemistry, and advanced screening technologies, these approaches enhance the identification of promising drug candidates. Overall, plant-derived natural products remain an essential foundation for modern pharmaceutical research, offering a diverse and sustainable source of novel bioactive molecules for the development of safer and more effective therapeutic agents.

Overview of *Limnophila Polystachya*

Limnophila polystachya Benth belongs to the family Scrophulariaceae is an herb and rare trees, mostly autotrophic and less often hemiparasitic. Stipules are absent; leaves are alternate, opposite, whorled or basally opposite and apically alternate (Zipcode Zoo.com). The plant species are widely used in traditional Indian medicine to treat pestilent fever, dysentery, elephantiasis, dyspepsia, antipyretic, expectorant, lactagogue and also exhibits significant antibacterial antifungal and antineoplastic activities 12. The *Limnophila polystachya* has been reported for its antitumor promoting agent, anti mutagenic 13, antimycobacterial and antioxidant activity.

TAXONOMY AND BOTANICAL CLASSIFICATION

Limnophila polystachya Benth. is a medicinal herb belonging to the genus *Limnophila* within the order Lamiales. Traditionally, it has been classified under the family Scrophulariaceae; however, according to the latest Angiosperm Phylogeny Group (APG IV) classification, the genus is now placed in the family Plantaginaceae. It is an angiosperm (Division: Magnoliophyta) and a dicotyledonous plant (Class: Magnoliopsida) belonging to the kingdom Plantae. The species is a semi-aquatic, aromatic annual or perennial herb commonly found in marshes, wetlands, paddy fields, and other waterlogged habitats. It is characterized by erect or creeping branched stems, opposite or whorled lanceolate leaves, and small bluish-purple flowers arranged in dense spikes. The fruit is a capsule containing numerous small seeds. *L. polystachya* is widely distributed throughout tropical and subtropical regions of South and

Southeast Asia, including India, Bangladesh, Nepal, Sri Lanka, Myanmar, Thailand, Malaysia, and Vietnam. Owing to its broad geographical distribution and traditional medicinal importance, the plant has attracted increasing scientific interest for its phytochemical composition and diverse pharmacological activities.

Phytoconstituents

Phytochemical and pharmacological studies on *Limnophila polystachya* remain limited, with very few investigations reporting its chemical constituents or biological activities. Consequently, its therapeutic potential has not been fully explored. However, studies on related *Limnophila* species have identified several bioactive compounds, including flavonoids such as 5-hydroxy-6,8-dimethoxy-3',4'-methylenedioxyflavone and 5,8-dihydroxy-6,7,4'-trimethoxyflavone from *Limnophila indica*. Additionally, essential oils have been isolated from *Limnophila rugosa*, and two novel flavonoids have also been reported from *L. indica*. These findings suggest that *L. polystachya* may also contain valuable phytochemicals with significant pharmacological potential, highlighting the need for further phytochemical and biological investigations.

MATERIALS AND METHODS

In the reported study, *Limnophila polystachya* Benth. leaves were collected from Samayapuram, Tiruchirappalli, Tamil Nadu, India, and authenticated by the Botanical Survey of India. The shade-dried leaves were powdered and extracted with ethanol using a cold maceration method for 14 days, followed by filtration and solvent evaporation to obtain the crude extract. The pharmacological activities of the extract were evaluated using Swiss albino mice under standard laboratory conditions and approved ethical guidelines.

The analgesic activity was assessed using the acetic acid-induced writhing model, where the ethanolic leaf extract was administered orally at doses of 250 and 500 mg/kg and compared with diclofenac sodium (10 mg/kg) as the standard drug. Antidiarrhoeal activity was evaluated using the castor oil-induced diarrhoea model, with loperamide (3 mg/kg) serving as the reference standard. Antimicrobial activity was determined by the disc diffusion method against four bacterial strains, including the Gram-positive bacteria *Bacillus subtilis* and *Staphylococcus aureus*, and the Gram-negative bacteria *Escherichia coli* and *Klebsiella oxytoca*. The antibacterial efficacy was assessed by measuring the diameter of the inhibition zones produced by the ethanolic leaf extract in comparison with the standard antibiotic

ciprofloxacin. These well-established experimental models enabled the evaluation of the analgesic, antidiarrhoeal, and antimicrobial potential of *L. polystachya* leaf extract.

Extraction methods

The ethanolic leaf extract of *Limnophila polystachya* was prepared by the cold maceration method. Approximately 300 g of shade-dried, powdered leaves were soaked in 1000 mL of ethanol for 14 days with intermittent shaking and stirring to ensure efficient extraction of phytoconstituents. The mixture was then filtered successively through cotton wool and Whatman filter paper to remove plant debris. Finally, the filtrate was concentrated under reduced pressure using a rotary evaporator to remove the solvent, yielding the crude ethanolic extract for subsequent pharmacological evaluation.

Pharmacological effects

Limnophila species possess a wide range of pharmacological activities, including antimicrobial, antidiarrhoeal, anti-inflammatory, antioxidant, antitubercular, wound-healing, cytotoxic, and anthelmintic effects. These activities are primarily attributed to bioactive compounds such as flavonoids (notably nevadensin and isothymusin), terpenoids, and essential oils. Experimental studies have demonstrated promising efficacy against various pathogens, inflammatory conditions, diarrhoeal diseases, and oxidative stress, while also supporting their traditional medicinal use. Although the available preclinical evidence is encouraging, further clinical studies are required to establish the safety and therapeutic efficacy of *Limnophila* species for pharmaceutical applications.

Toxicological evaluation

Toxicological studies have demonstrated that the ethanol leaf extract of *Limnophila polystachya* possesses a favorable safety profile. Acute toxicity evaluations in experimental rodents revealed no significant toxic manifestations or mortality following oral administration at doses up to 500 mg/kg. These findings suggest that the extract has a wide margin of safety and support its traditional medicinal use. Nevertheless, further sub-chronic, chronic, and clinical toxicity studies are required to comprehensively establish its long-term safety and therapeutic applicability.

Comparative analysis with other *Limnophila* species

Limnophila aromatica, *L. indica*, and *L. rugosa* share a similar phytochemical composition, being rich in flavonoids, phenolic compounds, and terpenoids, which are responsible for their

antimicrobial, antioxidant, and anti-inflammatory activities. Despite these similarities, each species contains distinct bioactive constituents that contribute to its unique pharmacological properties. *L. aromatica* is characterized by monoterpenes, phenylpropanoids, and polymethoxyflavones such as gardenin B and isothymusin, contributing to its antioxidant and vascular protective effects. *L. indica* contains unique trimethoxyflavones and phytosterols and is traditionally used for the treatment of diarrhoea, dysentery, and febrile illnesses. In contrast, *L. rugosa* is rich in nevadensin, salvigenin, betulin, and betulinic acid, which are associated with potent antimicrobial, anti-inflammatory, diuretic, and hypotensive activities. Although these species possess a common phytochemical framework, variations in their major constituents account for their distinct therapeutic potentials and traditional medicinal applications.

Limitations and future perspectives

Current research on *Limnophila polystachya* has demonstrated promising pharmacological activities, particularly antimicrobial and analgesic effects; however, several limitations remain. Most studies have focused on crude or partially purified extracts rather than isolated bioactive compounds, making it difficult to identify the exact constituents responsible for the observed biological activities. In addition, variations in phytochemical composition due to geographical origin, harvesting season, and extraction methods have limited the reproducibility and standardization of experimental findings. Furthermore, comprehensive investigations on pharmacokinetics, mechanisms of action, and long-term toxicity are still lacking, and the available evidence is largely confined to in vitro and animal studies, with no clinical validation in humans.

Future research should prioritize bioactivity-guided isolation and characterization of active phytoconstituents using advanced metabolomic approaches. The development of standardized herbal formulations and novel drug delivery systems, such as nanoformulations, may improve the bioavailability and therapeutic efficacy of the plant. In addition, detailed toxicological evaluations, pharmacokinetic studies, and well-designed clinical trials are essential to establish the safety, efficacy, and therapeutic potential of *L. polystachya*. Sustainable cultivation and conservation strategies should also be encouraged to ensure the long-term availability of this medicinal plant.

CONCLUSION

Overall, the available evidence suggests that the ethanol leaf extract of *Limnophila polystachya* possesses a favorable safety profile and exhibits significant antidiarrhoeal, antioxidant, and antimicrobial activities. These findings support its traditional ethnomedicinal use and highlight its potential as a promising source of novel therapeutic agents. Furthermore, the plant holds considerable potential for large-scale cultivation and commercialization, which may contribute to both pharmaceutical development and the socio-economic welfare of local communities.

REFERENCES

1. Jose BE, Jebaseelan S, Meera R and Kalirajan R: Analgesic, antidiarrhoeal and antimicrobial activities of *Limnophila polystachya* Benth (Scrophulariaceae) leaf extracts. *Int J Pharmacognosy* 2022; 9(6): 127-31.
doi link: [http://dx.doi.org/10.13040/IJPSR.0975-8232.IJP.9\(6\).127-31](http://dx.doi.org/10.13040/IJPSR.0975-8232.IJP.9(6).127-31).
2. View of Chemical and pharmacological aspects of *Limnophila geoffrayi*: An update. (n.d.). <https://wjpsonline.com/index.php/wjps/article/view/chemical-pharmacological-limnophila-geoffrayi-review/835>
3. Chaachouay, N., & Zidane, L. (2024). Plant-Derived Natural Products: A Source for Drug Discovery and Development. *Drugs and Drug Candidates*, 3(1), 184-207. <https://doi.org/10.3390/ddc3010011>
4. Brahmachari, G. (2014). *Limnophila* (Scrophulariaceae): Chemical and pharmaceutical aspects - an update. In *Bentham Open, The Open Natural Products Journal* (Vol. 7, pp. 1–14). <https://www.benthamopenarchives.com/contents/pdf/TONPJ/TONPJ-7-1.pdf>
5. Brahmachari, Goutam. (2015). *Limnophila* (Scrophulariaceae): Chemical and Pharmaceutical Aspects - An Update. *The Open Natural Products Journal*. 9. 1-14. 10.2174/1874848101407010001.